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The early life environment influences trajectory of post-partum weight loss

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CEA designed the study, performed experiments, drafted and edited the manuscript. JLA and TA performed experiments and edited the manuscipt. SEO designed the study and

edited the manuscript. The authors have all approved the final version of the manuscript.

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Abstract (255)

Research question: The physiological processes of pregnancy and lactation require profound changes in maternal metabolism and energy balance. The timescale of metabolic reversion after pregnancy, in particular postpartum weight loss, is highly variable between individuals. Currently, mechanisms influencing postpartum metabolic recovery are not well understood. We hypothesize that, in common with other metabolic and obesity-related outcomes, capacity for postpartum weight-loss is influenced by developmental programming.

Design: Adult female Wistar rats exposed to a maternal low-protein diet *in utero* then weaned onto a control diet postnatally (recuperated) were compared to controls. Adult females from both groups underwent pregnancy at 3 months of age. Weight changes and metabolic parameters during pregnancy and lactation were compared between control and recuperated groups, and also to non-pregnant littermates.

Results: Pregnancy weight gain was not different between the control and recuperated groups, but postpartum recuperated animals remained significantly heavier than both postpartum control animals ($p<0.05$) and their non-pregnant recuperated littermates ($p<0.05$) at the end of lactation. Postpartum recuperated animals had increased intra-

abdominal fat mass ($p<0.05$) and increased serum triglycerides ($p<0.01$) compared to controls. Postpartum recuperated animals also had increased expression of IL6, NRF2, and ALOX12 (key regulators of inflammation and lipoxygenase activity) in the intra-abdominal adipose tissue compared to control groups.

Conclusions: Mothers who themselves have been exposed to adverse early-life environments are likely to have slower metabolic recovery from pregnancy than controls. Failure to return to pre-pregnancy weight after delivery predisposes to persisting sequential inter-pregnancy weight gain, which can represent a significant metabolic burden across a life-course involving several pregnancies.

Key message:

Exposure to low-protein diet *in utero* influences post-partum metabolic recovery in female adult rats.

Key words: post-partum weight; developmental programming; adipose mass; metabolism; inter-pregnancy weight gain; rat; animal model; ovarian reserve; primordial follicles; reproductive ageing

Introduction

Healthy pregnancy requires major adaptations of maternal physiology to accommodate the needs of a growing fetus and to amass sufficient maternal energy reserves for delivery and lactation (Tan and Tan 2013). Key metabolic adaptations during pregnancy include increased adipose mass, reduced insulin sensitivity, increased propensity to fasting ketosis, and changes in body fluid composition (Lowe and Karban 2014, Lacroix et al. 2016, Staelens et al. 2016). A significant body of evidence suggests that an individual's response to metabolic challenges of pregnancy can be viewed as a 'stress test' that reveal the individual's underlying propensity to metabolic dysfunction (Williams 2003). For example, if the physiological reduction in normal glucose tolerance (due to placenta-derived insulin-resistance promoting peptides) provokes frank gestational diabetes, then the chance of developing type 2 diabetes in later life is at least 7-fold higher than in women who were normo-glycaemic in pregnancy (Bellamy et al. 2009). Thus the metabolic response to pregnancy can be viewed as both a window to future health, and an important opportunity to intervene to improve health over the life-course.

Over the course of a lifetime, failure to recover to baseline from the physiological challenges of each pregnancy can lead to gradual accumulation of fat mass and subsequent long-term metabolic derangement (Amorim et al. 2007, Lipsky et al. 2012). This cycle of inter-gestational weight gain leads to an increasingly higher BMI with each successive pregnancy, to the detriment of both the mother (Abrams et al. 2017) and potentially her subsequent offspring (Oostvogels et al. 2014). Evidence suggests that around half of women have regained their pre-pregnancy weight by 12 months postpartum (Sagedal et al. 2017) although this is highly dependent on factors such as maternal age, socio-economic status, pre-pregnancy BMI, stress, and breast-feeding (Jarlenski et al. 2014, Endres et al. 2015, Straub et al. 2016). Trials in human pregnancy show that diet and exercise interventions can be effective in limiting gestational weight gain (Muktabhant et al. 2015) and that these interventions may also be partially effective in promoting postpartum weight loss and altering maternal dietary behaviour (Ehrlich et al. 2014, Horan et al. 2014, Patel et al. 2017, Sagedal et al. 2017). However, very little is known about the molecular mechanisms that influence the propensity to post-partum weight loss in individuals.

Risk of obesity in adult life is increased by exposure to a suboptimal intrauterine environment (Bouret et al. 2015). We have previously demonstrated using a rat model that adult offspring exposed to low protein maternal diet during pregnancy followed by postnatal catch-up growth (recuperated) are prone to later life obesity (Cripps et al. 2009, Berends et al. 2013) and metabolic derangement (Martin-Gronert et al. 2008). We therefore hypothesized that an adult female who was exposed to a suboptimal early life environment (with resulting programmed energy and glucose handling deficits) might have reduced capacity for metabolic recovery postpartum in addition to an increased risk of obesity in later adulthood, compared to control counterparts..

The aetiology of post-partum adipose mass retention might also be explored by examining adipocyte gene expression in the immediate post-weaning period. Gene expression of key metabolic, endocrine and reproductive pathways may provide insight into the molecular pathways that regulate post-partum weight loss. In addition to quantifying intra-abdominal fat mass in the post-partum period, we also aimed to assay the relative expression levels of a panel of key candidate genes, including those involved in the inflammatory adipose response (*Il1, Il6, Tnfa, Il10*), oxidative stress response (*Hmox1, Xo, NfkB, Gp91phox*), lipoxygenase activity (*Alox12, Alox15*), macrophage infiltration (*Mcp1, Cd68*), and master transcriptional regulators thought to be involved in the pathogenesis of obesity (*Tgfb, Tnfa, Nrf2*). These genes were chosen based on (i) previous work on the effects of developmental programming on ovarian and adipose gene expression (Aiken et al. 2016) (ii) knowledge of programming mechanisms in other organ systems in the same recuperated animal model (Aiken et al. 2016, Tarry-Adkins et al. 2016) and (iii) relevant literature review based on searching the Pubmed and Medline databases using the MeSH terms: “Postpartum period” AND “Body Weight Change”.

We have previously shown a significant relationship between developmentally programmed obesity and reduced primordial follicular reserve (Aiken et al. 2016) in adult females. It is not currently known whether this is a result of reduced follicular endowment in the perinatal period or an accelerated decline in follicular reserve during reproductive life. Understanding whether primordial

follicular reserve is further reduced after the metabolic and endocrine challenge of pregnancy and lactation could give important insight into the dynamics of reduced ovarian reserve and hence future reproductive potential in developmentally-programmed animals.

Our aim in this study was therefore to determine whether postpartum weight loss and metabolic recovery from pregnancy was significantly different between control animals and those with programmed energy and glucose handling deficits resulting from exposure to a suboptimal intrauterine environment (recuperated group).

Materials and methods

Experimental Design

All animal experiments were approved by the University of Cambridge Animal Welfare and Ethical Review Board. All animal experiments were conducted in accordance with the British Animals (Scientific Procedures) Act (1986) and were compliant with EU Directive 2010/63/EU. The aim of the study was to test whether post-partum metabolic recovery and weight loss was impaired in adult females that had been exposed to a maternal low-protein diet *in utero*. Wistar rat dams (F0 generation, n=16) were fed a standard laboratory chow diet (20% protein) and fed *ad libitum* until pregnancy was confirmed through the observation of vaginal plugs. Pregnant animals were then randomly assigned to a 20% protein diet (control) or an 8% isocaloric low protein (LP) diet (n=8 in each group), as described previously (Martin-Gronert et al. 2008). Both diets were purchased from Arie Blok (Woerden, The Netherlands). Pups born to LP-diet-fed dams were cross-fostered to control-fed mothers at postnatal day 3, in order to create recuperated offspring. Each recuperated litter was culled to 4 female pups (F1 generation, recuperated group, n=8 litters) at random to maximize their plane of nutrition. The control group was the offspring of mothers fed the 20% protein diet during gestation and suckled by 20%-protein-fed dams during lactation (F1 generation, control group, n=8 litters). Each control litter was culled to 8 female pups. After weaning, all first-generation offspring were

maintained on standard laboratory chow fed *ad libitum*. At 12 weeks of age, two first-generation females from each litter were randomly selected. One F1 female from each F0 litter (n=8 in each group) was paired with a stud male and the day of mating confirmed by the presence of a vaginal plug. The other female from each litter was not mated and maintained in standard conditions as described previously.

During pregnancy, serial body weights were obtained every 4 days from both the pregnant female and her non-pregnant littermate. After giving birth, the second-generation (F2) litters were all culled to 8 pups (both F2 control and F2 recuperated groups) on day 3 and were all suckled by their own mothers. At day 3 the first postnatal body weights were obtained, and thereafter every 7 days until weaning. There was no significant difference in birth-weight or litter size in the F2 offspring between experimental groups. Detailed phenotyping of the F2 generation of this cohort is described elsewhere (Aiken et al. 2015, Tarry-Adkins et al. 2018). At weaning of the second-generation litter (postnatal day 21), all first-generation females (n=8 post-partum mothers and their 8 non-pregnant littermates, in each of the control and recuperated groups) were fasted overnight and fasting blood glucose levels were determined using a glucometer (Hemocue, Angelholm, Sweden). The first-generation females were culled by carbon dioxide asphyxiation and cervical dislocation. At postmortem serum samples, ovaries, ovarian fat pads, and other solid intra-abdominal organs were harvested and weighed fresh, immediately after dissection. One ovary from each animal was snap-frozen in liquid nitrogen and the other fixed in formalin/paraldehyde. The fixed ovaries were sectioned and subjected to haematoxylin and eosin (H&E) staining to ensure equal distribution of estrous stages in each experimental group (data not shown). Sample analysis was performed using project codes to blind the investigators to the experimental groups. Seven samples per group were analyzed at each time-point, each sample representing a different litter. The sample size was determined via a power calculation based on the effect sizes seen in our previous studies (Aiken et al. 2013, Aiken et al. 2015), using an alpha level of 0.05 to give power of 0.8.

Serum analyte measurements

Blood was obtained from the tail vein into EDTA tubes and centrifuged for 3 min at 955xg at 4° Celsius to isolate serum. Fasted blood glucose measurements were obtained using a glucose analyzer (Hemocue, Angelholm, Sweden). The serum lipid profiles were performed using an auto-analyzer (MRC MDU Mouse Biochemistry Laboratory). Serum leptin was measured using an ELISA kit from Crystal Chem (Zaandam, The Netherlands), which was used according to the manufacturer's instructions.

Primordial follicle counts

Fixed ovaries were processed for microscopy and the entire ovary sectioned at 8µm. Every 9th section was stained with H&E for morphometric analysis (72µm between analysed sections). Only follicles with a visible oocyte nucleus were counted, in order to avoid repeat counts of the same follicle (Bernal et al. 2010). Primordial follicles were identified morphologically by the presence of a single layer of flattened granulosa cells surrounding the oocyte (Picut et al. 2014). Total volume of each ovary was calculated (section areas x section thickness x number of sections) and the follicle count expressed as follicles/mm^3 of ovarian tissue.

Gene expression analysis

A panel of 15 candidate genes was developed to test which molecular pathways might be involved in post-partum metabolic recovery. RNA was extracted from snap-frozen para-ovarian fat pads using a miRNeasy mini kit (Qiagen, Hilden, Germany) following manufacturers' instructions, with the addition of a DNaseI digestion step to ensure no genomic DNA contamination. RNA quantification was performed using a NanoDrop spectrophotometer (Nanodrop Technologies, Wilmington, DE, USA). RNA (1 µg) was used to synthesize cDNA using oligo-dT primers and M-MLV reverse transcriptase (Promega, Madison, Wisconsin, USA). Gene expression was determined using custom designed primers (Sigma, Poole, Dorset, UK) and SYBR Green reagents (Applied Biosystems, Warrington, UK) as previously described ((Tarry-Adkins et al. 2009). Quantification of gene expression was performed using a Step One Plus RT-PCR machine (Applied Biosystems, Warrington, UK). Equal efficiency of the reverse transcription of RNA from all groups was confirmed through quantification of expression of the house-keeping gene *ppia*, the expression of which did not differ between groups.

Statistical Analysis

All data were analyzed using hierarchical linear models with a random effect for litter of origin. Maternal diet and pregnancy status were included as fixed effects. This structure accounted for the fact that post-partum and non-pregnant littermates are derived from a single pregnancy, and these data are therefore effectively paired and cannot be treated as fully independent. Multiple hypothesis correction testing was performed using the p values obtained from the regression models, correcting for the false discovery rate (FDR). All bodyweights were expressed as a ratio of current weight/weight on study day 0. Data are represented as means ± SE. A value of $p < 0.05$ was considered statistically significant. All data analysis was conducted using the R statistical software package version 2.14.1 (R Foundation for Statistical Computing, Vienna, Austria). In all cases, n refers to the number of litters.

Results

Peripartum bodyweights

There was no significant difference in average weight gain during pregnancy in the control versus the recuperated group (Figure 1). At full term (day 20 after conception), the control and recuperated pregnant mothers were both approximately 40-50% heavier than their pre-pregnancy weights (control pregnant 1.38-fold ±0.06 versus recuperated pregnant 1.47-fold ±0.04). There was no difference in litter size or pup weight in the control versus the recuperated groups (full characterization of the F2 generation has been previously reported (Aiken et al. 2015)). However, by the end of lactation (day 45 post-conception and day 24 post-delivery), the recuperated postpartum mothers remained heavier than their control postpartum counterparts (control postpartum 1.13-fold ± 0.03 versus recuperated postpartum 1.24±0.01, $p<0.01$). Compared to their control non-pregnant littermates the control postpartum mothers were not significantly heavier (control postpartum 1.13-fold ± 0.02 *versus* control non-pregnant 1.15-fold ± 0.02), however the recuperated mothers were

significantly heavier than their recuperated non-pregnant littermates (recuperated postpartum 1.24 ± 0.01 versus recuperated non-pregnant 1.19 ± 0.01, $p<0.05$).

Organ weights

Para-ovarian fat pad weights were significantly higher in recuperated groups compared to controls ($p<0.05$), and non-significantly higher in the post-partum groups compared to the non-pregnant littermates ($p=0.07$) (Figure 2A). Overall, there was a significant interactive effect of recuperated status and post-partum status ($p<0.05$), indicating that intra-abdominal fat mass is more likely to be retained post-partum in recuperated animals. Uterine weights were not significantly different between any of the experimental groups (Figure 2B). This suggests that complete involution of the uterus had occurred by the time of study sampling, and thus the effects observed during the study were not the rapid dynamic changes of the early post-partum period, but a stable phenotype in the post-delivery phase.

Serum analytes

There was no difference in blood glucose or serum insulin levels between any of the experimental groups (Table 1). There was a trend towards lower leptin in the postpartum groups ($p=0.08$), which likely represents lactational hypoleptinaemia, but no significant effect of recuperated versus control status ($p=0.11$). The relative suppression of leptin during lactation is a previously characterized effect, which drives a chronic hyperphagia to meet the metabolic demands of milk production (Smith et al. 2010).

There was a significant rise in serum cholesterol in both post-partum groups compared to their non-pregnant littermates ($p<0.01$), but no effect of recuperated status (Figure 3A). Fasting serum triglycerides were elevated in both the post-partum groups ($p<0.01$) and in the recuperated maternal dietary groups ($p<0.01$) (Figure 3B). Fasting free fatty acids were higher in the recuperated groups than in the controls ($p<0.01$), but unaffected by pregnancy status.

Primordial follicle counts

Ovary weights were significantly higher in the postpartum groups than in the non-pregnant littermates ($p<0.001$), but there was no difference between recuperated and control groups (Figure 2C). Primordial follicle counts per cubic millimeter of ovarian tissue were significantly higher in the control than in the recuperated groups ($p<0.05$), an effect that has previously been described in this model at 6 months of age ((Aiken et al. 2013). There was no effect of having recently been pregnant on primordial follicle counts (Figure 4), which is to be expected, given the relatively short duration of the gestation period.

Screening for differences in para-ovarian fat pad gene expression

A candidate screen on 15 genes was developed, and the screening results were corrected for multiple hypothesis testing. There were no significant interactive effects between postpartum status and recuperated versus control status on expression levels of any of the candidate genes in para-ovarian adipose tissue (Table 2).

(i) Effect of recuperated versus control status on gene expression in para-ovarian adipose tissue

Recuperated versus control status in postpartum rats had a significant effect on expression levels of two candidate genes (Table 2). *Nrf2,* an important DNA-binding transcription factor that regulates mitochondrial biogenesis, was increased in recuperated adult females compared to controls ($p<0.05$). This is particularly interesting in light of other evidence that suggests *Nrf2* may be up-regulated in other maternal dietary models of developmental programming (Aiken et al. 2016). *Il6,* which is a major pro-inflammatory cytokine, was also up-regulated in adipose tissue in the recuperated group ($p<0.05$). This is important as it suggests not only is the total mass of intra-abdominal tissue increased, but that one of its major detrimental effects, i.e. producing a phenotype of chronic inflammatory response, may be exacerbated in this developmental programming model.

(ii) Effects of recent pregnancy on gene expression in para-ovarian adipose tissue

Expression of *Cd68*, a marker of monocyte lineage, was elevated in animals that had recently been pregnant ($p<0.05$) (Table 2), compared to their non-pregnant littermates. Increased monocyte infiltration following pregnancy suggests increased inflammation in the adipose tissue. Furthermore, there was a significant increase in $Gp91^{phox}$ expression in the postpartum group ($p<0.05$), which potentially reflects an increase in oxidative stress in the para-ovarian adipose tissue.

Discussion

In this study we show that post-partum weight loss is influenced by early life exposure to an undernourished intrauterine environment. It has previously been established in a variety of animal developmental programming models (Samuelsson et al. 2008, Samuelsson et al. 2013, Sun et al. 2014), and suggested by human epidemiological data (Finer et al. 2016, Mitanchez and Chavatte-Palmer 2018), that adult females who have been exposed to adverse early life environments, e.g. via suboptimal maternal diet, are more likely to become obese in later life. This study advances our understanding of how pregnancy and post-partum recovery can be an important factor in influencing the propensity to obesity in developmentally programmed females.

In our rodent model, suboptimal *in utero* nutrition significantly increased postpartum weight retention at the end of lactation. In particular, adult females exposed to a maternal low-protein diet *in utero* were on average 24% heavier than their own pre-pregnancy weights at the end of lactation (compared to 13% heavier in the control group). We demonstrated that developmentally programmed animals that undergo pregnancy have a prolonged exposure to increased intra-abdominal fat mass compared to controls, a conclusion that is reflected in both direct measurement of the intra-abdominal fat pad and in elevated concentrations of serum triglyceride compared to control postpartum animals. Increased visceral adiposity is known to be detrimental to long-term health via a variety of mechanisms including a chronic inflammatory response (Schlecht et al. 2016) which is in keeping with our observations of increased *Cd68* and *Il6* gene expression. However there was no evidence of any interaction between postpartum status and maternal diet in any particular gene expression pathway that could provide a clear insight into the molecular mechanism of postpartum weight

retention in the programmed animals. However, the observed elevation in free fatty acids in the recuperated group is in keeping with adipose tissue insulin resistance.

Over 80% of adult females in the UK experience a viable pregnancy during their lifetime (ONS 2016). Despite the fact that pregnancy is an extremely common life-event, the majority of female animals in developmental programming studies do not experience pregnancy as part of the longitudinal cohort structure. Even when animal models are bred to produce an F2 generation, it is rare that the postpartum outcomes of the F1 mothers are reported. However, it is increasingly understood that the physiological response to pregnancy and post-partum recovery can provide a useful window into later metabolic health (Drost et al. 2013, Visser et al. 2014). In common with gestational diabetes (Bellamy et al. 2009) and hypertension in pregnancy (Hermes et al. 2012), peripartum body-weight changes could be a useful parameter to help identify individuals at high risk of later weight gain and poor cardiovascular and metabolic health. In addition to enabling identification of individuals at risk of obesity, postpartum weight retention may be an important factor in the causal mechanism of obesity in adult females (Ketterl et al. 2018). Post-partum weight loss and its inverse effect, inter-gestational weight gain, are becoming increasingly recognized as major determinants of women's health across the life-course (Oteng-Ntim et al. 2018). A recent human study found that, in normal weight women who received standard pregnancy care, only 20.7% had regained their pre-pregnancy weight at 6 months post-partum, and that the average weight retention was 3.3 ± 3.5 kg (Phelan et al. 2011). Among those who had not returned to pre-pregnancy weight by 12 months, the average weight retention remained relatively stable at 3 ±5.7 kg (Phelan et al. 2014). Over the course of several full-term pregnancies, each additional weight gain can thus accumulate to produce a significant metabolic burden (Hutcheon et al. 2017) in the mother, with additional potential detrimental consequences for children born from subsequent pregnancies. It is thus an important facet of life-course health in women to understand as much as possible about factors that influence post-partum weight loss. It is not possible to say whether the findings from our rodent population will be directly applicable to human pregnancy cohorts, hence the need for further study of factors influencing post-partum weight loss in human pregnancy cohorts.

A major advantage of the study design was the ability to compare each animal with a non-pregnant littermate, which allowed us to control for observable and non-observable factors related to the early-life environment, and hence isolate specifically the implications of pregnancy. Furthermore, we were able to successfully control for the fact that these early-reproductive life animals were still themselves growing during the study period.

Limitations of the current study include the inability to follow these animals through a subsequent pregnancy and into reproductive senescence to measure whether post-partum weight loss directly influenced the extent of later life weight gain. Unfortunately this was beyond the scope of the design of the current cohort. Other limitations include the inability to vary the length of the lactation period, which may have a significant influence on postpartum weight loss or to study post-partum weight loss in mothers who did not suckle their offspring (which is common in human populations). This was not possible because of the need to standardize conditions for the F2 generation, whose outcomes have been reported elsewhere (Aiken et al. 2015).

In conclusion, we have shown that exposure to a suboptimal early life environment influences the rate of post-partum weight loss. Understanding the factors that make post-partum weight loss more difficult represents the first key step towards developing interventions that can improve the percentage of women who have regained their pre-pregnancy weight by the start of the next pregnancy, and hence reduce their chances of obesity over their lifetime.

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Table and Figure Legends:

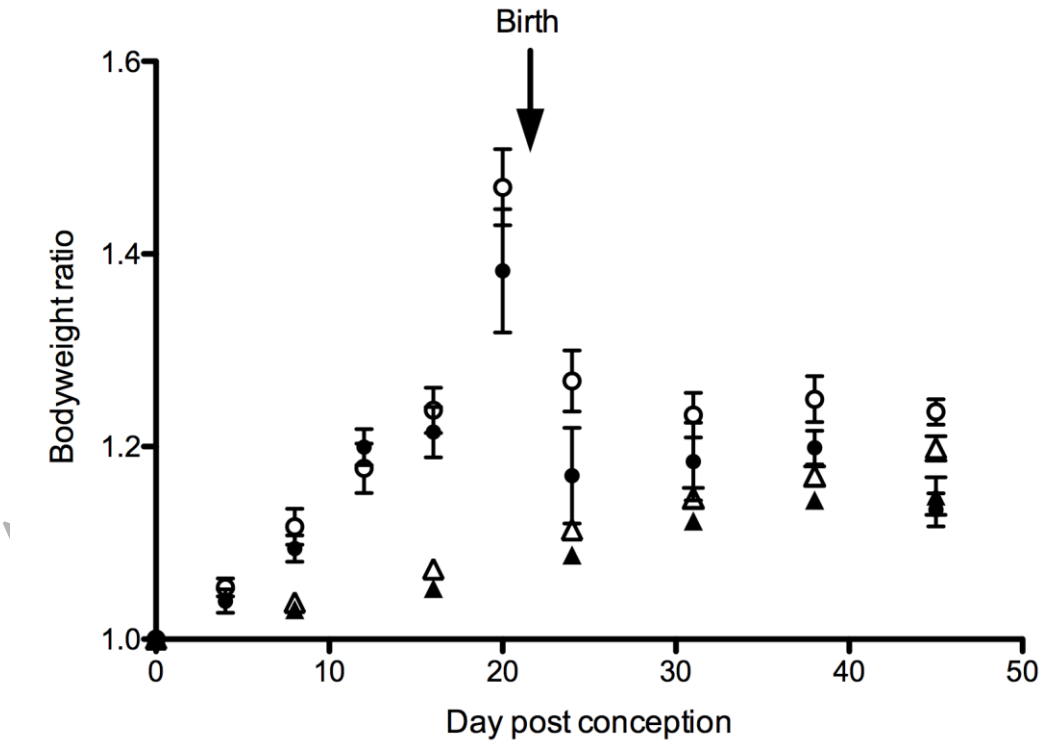

Figure 1 Body weights of adult females during pregnancy and lactation. All bodyweights are normalized to the weight at the start of the study. There was no significant

difference in starting body weights between groups. Open circles: pregnant recuperated group, Closed circles: pregnant control group, Open triangles: non-pregnant recuperated group, Closed triangles: non-pregnant control group. At the final study time point, the post-partum recuperated group was significantly heavier than any other study group.

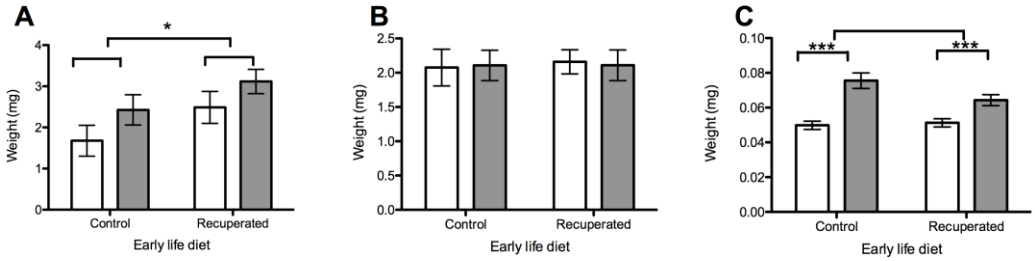

Figure 2 Organ weights in the postpartum period. Open bars: non-pregnant littermates, grey bars: post-partum females. A) Para-ovarian fat pad, B) Uterus weight, C) Ovary weight *p<0.05, ***p<0.001.

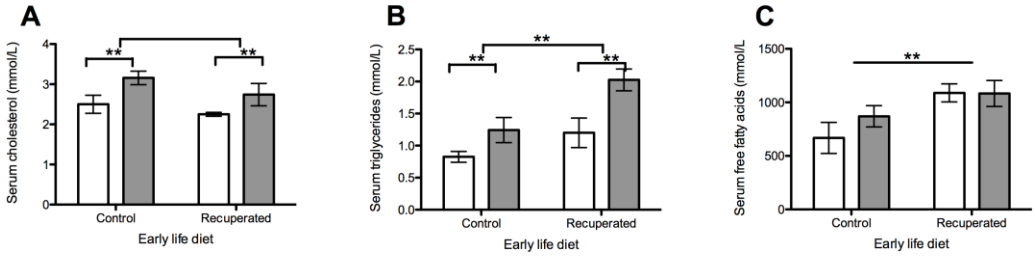

Figure 3 Maternal serum lipid profiles. All testing was performed following a period of overnight fasting. Open bars: non-pregnant littermates, grey bars: post-partum females. A) Serum cholesterol (mmol/L), B) Serum triglycerides (mmol/L), C) Serum free fatty acids (mmol/L). **p<0.01.

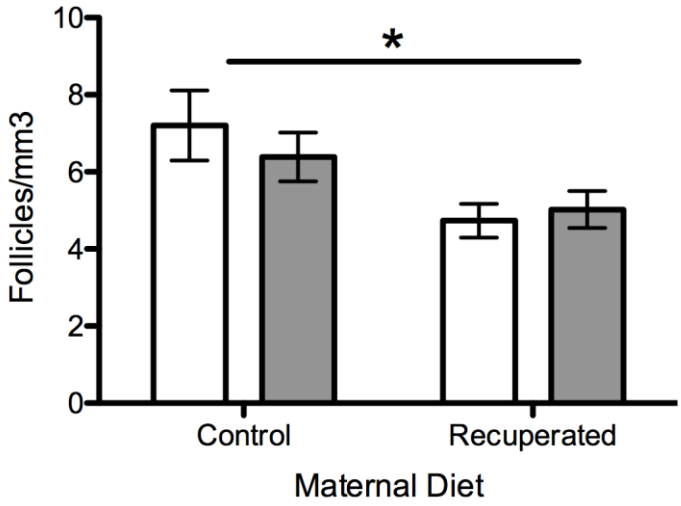

Figure 4 Primordial follicular reserve. Open bars: non-pregnant littermates, grey bars: post-partum females. Primordial follicular reserve was higher in the control than the recuperated groups, but there was no significant difference by postpartum status. *p<0.05.

Table 1 Levels of glucose, insulin and leptin in maternal serum by pregnancy and dietary group

	Control Postpartum	Control Non-pregnant	Recuperated Postpartum	Recuperated Non-pregnant	Effect of pregnancy	Effect of maternal diet
Blood glucose (mmol/L)	5.5 ± 0.4	5.3 ± 0.4	5.6 ± 0.3	5.4 ± 0.3	p=0.88	p=0.72
Serum insulin (ng/ml)	0.8 ± 0.04	0.8 ± 0.03	0.9 ± 0.06	0.8 ± 0.08	p=0.22	p=0.64
Serum leptin (ng/ml)	5.2 ± 0.71	7.1 ± 1.5	7.3 ± 0.1	10.5 ± 1.29	p=0.11	p=0.08

Table 2 Effect of pregnancy and maternal diet on gene expression in the para-ovarian fat pad of adult female rats. All reported p values have been adjusted to take account of multiple hypothesis testing. *p<0.05

Gene	Pregnancy effect	Maternal diet effect	Interaction effect
Ppia	0.22	0.23	0.60
Nrf2	0.01*	0.16	0.59
Hmox1	0.14	0.46	0.28
Xo	0.56	0.35	0.62
Alox12	0.51	0.32	0.05
NfkB	0.97	0.09	0.13
Il6	0.02*	0.58	0.65
I1b	0.46	0.24	0.36
Tnfa	0.57	0.29	0.83
Tgfb	0.78	0.40	0.50
Il10	0.22	0.23	0.60
Alox15	0.41	0.62	0.29
Mcp1	0.06	0.05	0.83
Cd68	0.77	0.03*	0.27
Gp91phox	0.87	0.03*	0.51

Author Biography

Dr Catherine Aiken is a University Lecturer in Maternal and Fetal Medicine at the University of Cambridge. Her research interests focus on the long-term outcomes of high-risk pregnancies for both mothers and infants, in particular metabolic outcomes. She is a practicing clinician and academic researcher.